

## **NON-TECHNICAL ABSTRACT**

Survival for children with advanced stage neuroblastoma, sarcoma, and retinoblastoma is poor in spite of intensive multiagent regimens. High-dose regimens with bone marrow or stem cell rescue have shown some promise in neuroblastoma and are being tested in sarcomas and retinoblastoma. Studies here at St. Jude Children's Research Hospital support the contention that tumor contamination of the stem cell product infused following high dose chemotherapy can contribute to relapse, and that therefore removal of residual tumor cells from the hematopoietic graft may be essential to improving patient outcomes. To address these issues, we propose to conduct a novel phase II purging safety study in children with metastatic neuroblastoma, sarcoma or retinoblastoma. This study will measure the safety and time to engraftment following CD34+ selection using a CD34+ antibody column (Isolex 300/ stem cell selection system) followed by selection by size and side scatter characteristics using a FACS Vantage high speed cell sorter.

We then propose to divide the purged stem cell product into two aliquots to be marked with similar recombinant retroviral vectors under different conditions. Retroviral marking provides a sensitive assay for the presence of small numbers of contaminating tumor cells in clinical autografts, and an assay for the source of cells contributing to relapse. Marking also provides a means to study engraftment of the manipulated autologous cells. Without marking, it is impossible to determine if reconstitution of hematopoiesis is due to autologous reinfused cells or due to recovery of endogenous stem cells that survived the transplant conditioning regimen. The source of reconstitution (graft versus endogenous) is particularly important in this study to determine if the ex vivo manipulations required for purging are adversely affecting the engraftment capacity of the harvested CD34+ cells. The dual marking component of the study will allow us to study conditions designed to achieve efficient transduction of reconstituting hematopoietic stem cells. Derivation of such conditions would be of benefit to a wide variety of patients who could potentially be treated with gene therapy approaches.